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Face-based perception of emotions in dairy goats

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Abstract

Faces of conspecifics convey information about identity, but also gaze, and attentional or emotional state. As a cognitive process, face-based emotion recognition can be subject to judgment bias. In this study we investigated whether dairy goats (n=32) would show different responses to 2-D images of faces of familiar conspecifics displaying positive or negative emotional states. We also examined the possible use of images of faces as stimuli in cognitive bias studies. The faces of four subjects were photographed in a positive and a negative situation. Three types of images of ambiguous facial expressions were then created using morphing software (75% positive, 50% positive, and 25% positive). In a test-pen, each goat was exposed for 3 seconds to each type of image, obtained from the same goat. All goats were shown non-morphed faces first, before being shown the three types of morphed faces, balanced for order. Finally, the first non-morphed face was shown again. Spontaneous behavioural reactions including ear postures (forward, backward and asymmetrical) and interactions with the screen (time spent looking or touching) were recorded during the 3 seconds. Results were analysed using REML with repeated measurements. Goats spent more time with their ears forward when the negative was shown compared to the positive ($F_{4,121.3}$ = 2.51, P = 0.018), indicating greater interest in negative faces. Identity of the photographed goat influenced the time spent with the ears forward ($F_{2,57.4}$ = 7.01, P = 0.002). We conclude that goats react differently to images of faces displaying different emotional states and that they seem to perceive the emotional valence expressed in these images. Response to morphed faces was not necessarily intermediate to response to negative and positive faces, and not on a continuum. Further study is thus needed to clarify the potential use of faces in cognitive bias studies.

Key words: goats, face, emotions, cognitive bias, ear postures

1 Introduction

It is now generally accepted both in the scientific community and by policy makers that animals are sentient beings, capable of experiencing emotions (de Vere and Kuczaj, 2016). Being able to assess emotional states in farm animals is crucial to improving their welfare. Emotions are defined as short-term internal psychological states induced by stimuli. According to Dantzer (2002) an emotional state has behavioural (e.g. running away from a frightening stimulus), physiological component (e.g. increase of heart-rate) and subjective (e.g. 'I feel frightened') components. Evidence of behavioural and physiological components of emotions has been shown repeatedly in animals (Désiré et al., 2002). The subjective dimension of emotions is of course difficult to evaluate in animals, since there can be no use of language for self-report as in psychology. However the development of methodologies such as judgement bias or attention bias tests in animals can give the researcher an indirect access to the subjective dimension of emotions in animals (Roelofs et al., 2016). An emotion can also be characterised by a combination of its valence, i.e. positive vs. negative, and its arousal, i.e. low or high. For example, fear has a negative valence and a high level of arousal (Mendl et al., 2010).

Although the function of emotion is not primarily for communication, the outward expression of an emotional state involves changes in posture, vocalisations, odours and facial expressions, which can be perceived and used as indicators of emotional state by other animals (Siniscalchi et al., 2013; Terlouw et al., 1998). Since conspecifics can perceive one another's emotions, understanding how emotions are identified and how they can spread within a social group could have a major impact on improving the welfare of farmed species that are reared in groups. This study was a step in that direction and focused on face-based emotion recognition in goats. The fact that the facial expressions of humans and nonhuman mammals have a lot in common was suggested first by Darwin (1872). For social species, faces are a major source of information (Little et al., 2011); features that allow the identification of the individual, but also the direction of gaze, attentional state and emotional state are conveyed through the face (Adolphs, 2002). Face perception, and more specifically the processing of emotions, has been widely studied in sheep, which can discriminate between calm and stressed faces of conspecifics and humans in 2-D images (Tate et al., 2006).

As small ruminants, goats are closely related to sheep. We therefore hypothesised that face-based perception of emotions in goats would be as developed as in sheep. Since goats display behavioural expressions that differ between situations of positive and negative valence (Briefer et al., 2015), we wished to determine if those displays would impact the goats' faces sufficiently so that a difference could be perceived by conspecifics. We therefore tested whether goats would react differently to 2-D images of faces of familiar conspecifics displaying positive or negative emotional states. The images used were obtained by filming goats during two types of interactions with a human handler. We also hypothesised that goats would display behaviours indicating negative valence when looking at negative faces, and positive valence when looking at positive ones.

Recent studies demonstrated that the emotional state of an animal can influence cognitive processes, such as learning, attention or judgement (Mendl et al., 2009). Judgement bias tests have been used in farm animals to assess emotional states, especially after manipulation of the environment to induce positive or negative emotional states or as a tool to assess the impact of husbandry practices (reviewed by Baciadonna and McElligott, 2015). Animals in a negative emotional state show pessimistic judgements (i.e., react in a similar way to negative and ambiguous stimuli) while those in a positive emotional state make optimistic judgements about ambiguous stimuli (i.e., react in a similar way to positive and ambiguous stimuli). Face-based perception of emotion is a cognitive process (Martin et al., 2012) and as such is potentially subject to this judgement bias. To test if images of faces could be used as cognitive bias stimuli, we produced three types of ambiguous faces ranging in valence from negative, using morphing software. For these images to be usable in cognitive bias studies, goats have to show distinct spontaneous reactions to images of goat faces taken in positive or negative situations. Furthermore goats have to show gradual intermediate responses to the morphed faces to comply with the cognitive bias response pattern.

Finally, since goats were exposed repeatedly and without reinforcement to the same type of stimuli, we wanted to test their level of attention after five exposures, and thus included a final test session that was a repeat of the first.

2 Methods

2.1 Ethical note

All experimental procedures were approved by the Animal Welfare Advisory Board of the research unit (INRA) and complied with the GRICE (Groupe de réflexion interprofessionnel sur les comités d'éthique appliquée à l'expérimentation animale) recommendations.

2.2 Animals and management

The experimental work took place between April and May 2015 at the INRA experimental farm at Thiverval-Grignon, France. 32 lactating Saanen (n=17) and Alpine (n=15) goats aged 18 months were used in this experiment. The animals had been removed from their dams after birth and artificially reared in mixed-breed groups. They were all familiar with each other, having lived in the same group for at least six months prior to the trial.

The 32 goats were tested in two groups of 16, balanced for breed (Group 1: 8 Alpine and 8 Saanen ; Group 2: 7 Alpine and 9 Saanen) and weight (Group 1: 55.3 ± 6.5 kg ; Group 2: 53.7 ± 5.2 kg). For the duration of the study, goats from both groups were housed together in the same straw pen that was set within the main farm building. Morning milking took place between 07.30 and 09.30, and afternoon milking between 15.30 and 17.30. The goats were fed a total mixed ration twice a day ad libitum. Goats had unlimited access to water.

For each group the tests were completed in four days. Two days separated the trials for Group 1 and Group 2.

2.3 Images of faces

Amongst the 32 goats, two Saanen and two Alpine were selected to be filmed to produce images of faces. The choice of the filmed animals, hereafter referred to as Photo Goats, was based on their individual reactions to humans. Since the positive situation consisted of a positive interaction with an experimenter, the first two goats of each breed to approach the experimenter of their own volition in the home pen were selected to be the Photo Goats. To produce the images, each Photo Goat was placed into two different situations that were likely to elicit a positive and a negative emotional state respectively. Rewarding stimuli are thought to elicit positive emotional states, while fitness-threatening stimuli (predator, pain, stress) elicit negative emotional states (Mendl et al., 2010). Behavioural observations were used in conjunction with this framework to determine the valence of the situation the goats were placed in.

Photo Goat faces were filmed with a HD camera (HDR-XR155, Sony, Japan). Frames with a full clear frontal view of the face were extracted from those short video clips using Pinnacle Studio 17 (Pinnacle Systems, 2013). The faces were then digitally cut from the frames and placed against a neutral beige background (RGB model: R=217, G=202, B=126) with Adobe Photoshop CC (Adobe Systems, 2014) to create the images used in the tests (Figure 1).

2.3.1 Positive situation

The Photo Goats were groomed by a familiar experimenter in the home pen. Pleasant grooming consisted of gentle scratching of the neck and shoulder areas for approximately 5 minutes. Grooming of this sort has been shown to be a gentle interaction in cattle (Schmied et al., 2008) and to induce a positive judgment bias in goats (Baciadonna et al., 2016). Since the Photo Goats had been chosen based on the fact that they voluntarily approached humans, habituation was not necessary. During grooming the Photo Goats did not move away and after grooming they repeatedly sought attention from the experimenter. These observations supported the idea that grooming was pleasurable and thus rewarding and induced an emotional state of low arousal and positive valence (Coulon et al., 2015). Goats had their ears lowered and turned down during almost the entire grooming session, and pictures of the animal displaying this ear posture were extracted from the videos. These images are hereafter named the positive images (Figure1).

2.3.2 Negative situation

Each Photo Goat was isolated in a weigh-crate, located within the main building, thus allowing continued auditory and olfactory contact with other goats. The negative stimulus was produced by an experimenter who applied an ice block to the udder for a maximum of 30 seconds, or until a negative reaction from the goat (e.g. stamping, sharp head movements, trying to leave the crate) was observed. The obvious thermal discomfort induced by the application of the ice pack made Ice a fitness-threatening situation. This is highlighted by attempts made by the Photo Goat to escape the source of discomfort and the situation was thus considered to have induced a negative state of high arousal. As soon as a good quality video was captured the goat was brought back to the group. All Photo Goats displayed a negative reaction and avoidance behaviours when the ice block was applied, which suggests that it did elicit an emotional state of high arousal and negative valence.

Pictures from the first reaction of the goat to the ice block were extracted from the films, when the animal raised its head, with the tip of the ears pointing backward and the auricles turned downwards. These images are hereafter named the negative images (Figure 1).

2.3.3 Morphed faces

The use of morphed images allowed the creation of intermediate images that were 25% (I-), 50% (I50) and 75% (I+) between the negative and positive images (Figure 1).

Intermediate stimuli of each Photo Goat were produced by morphing images obtained in a negative and in a positive situation from the same goat (WinMorph 3.01, DebugMode, 2012). Key facial-features such as eyes, nostrils, mouth, ears and shape of the forehead and the jaw were marked on the positive and negative faces (Figure 1a). The positive face was then distorted into the negative one frame by frame.

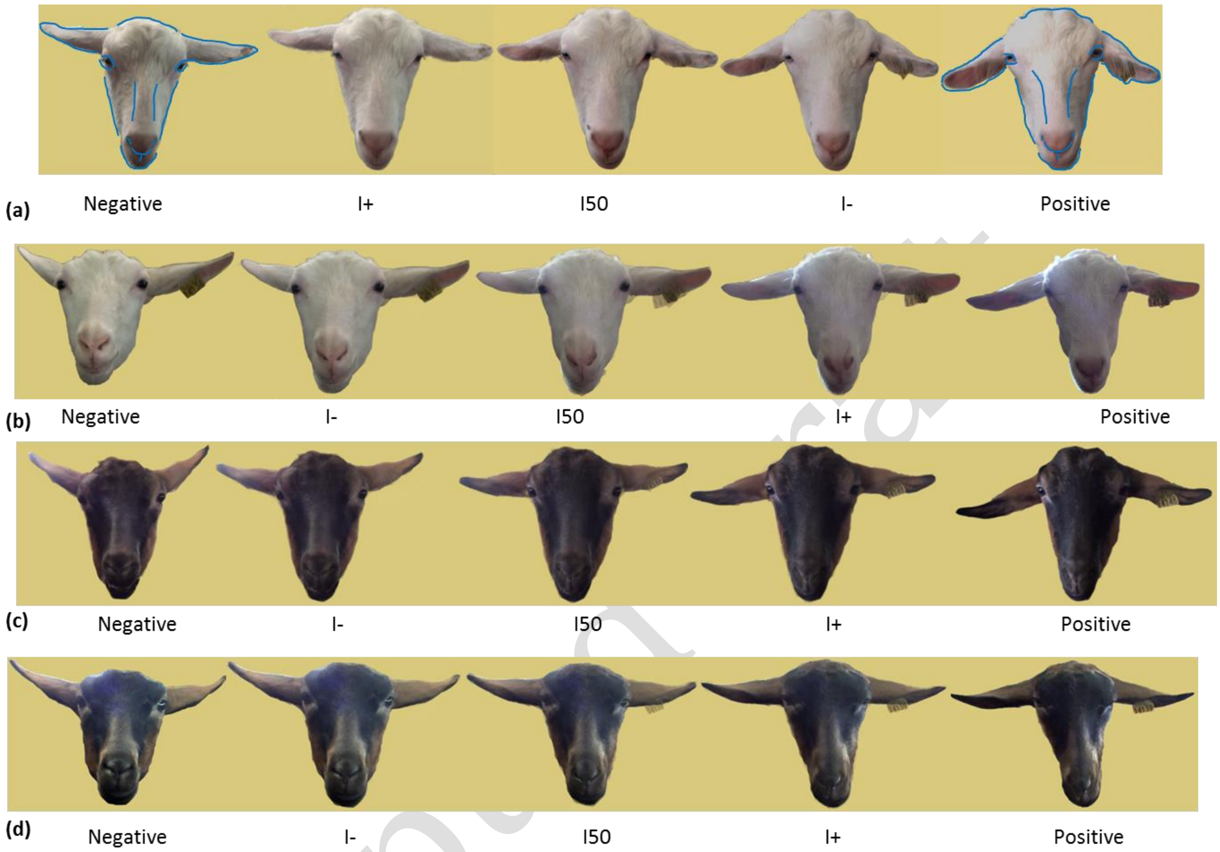

Figure 1.The five types of images of faces obtained from four different goats: (a) Photo Goat Saanen 1 (b) Photo Goat Saanen 2, (c) Photo Goat Alpine 1, (d) Photo Goat Alpine 2. 'Negative' images were taken when an icepack was applied to the udder. 'Positive' images were taken while the goat was being groomed by a familiar experimenter. The three types of images of ambiguous facial expressions were created using morphing software (25% positive (I-), 50% positive (I50), and 75% positive (I+)). The blue lines (a) outline key facial-features marked on the positive and negative faces in the morphing software.

2.4 Tests: spontaneous reactions to images of faces

2.4.1 Test pen

The test pen was located outside the main building in a covered area approximately 40m away from the home pen (Figure 2). The waiting pen was adjacent to the test pen but separated from it by a wall of straw bales. The test pen had solid wooden walls. An extra wooden panel prevented entry to one corner of the test pen and prevented goats from standing in the blind spot of the cameras. A computer screen (19 inch, Dell) was placed on the wall opposite the entrance at eye-level for goats, to display images of faces. The screen was not present during the habituation period. There was a small opening in the solid wall above the screen. During test sessions, the experimenter could place small items through the hole to draw the attention of the goat to the computer screen.

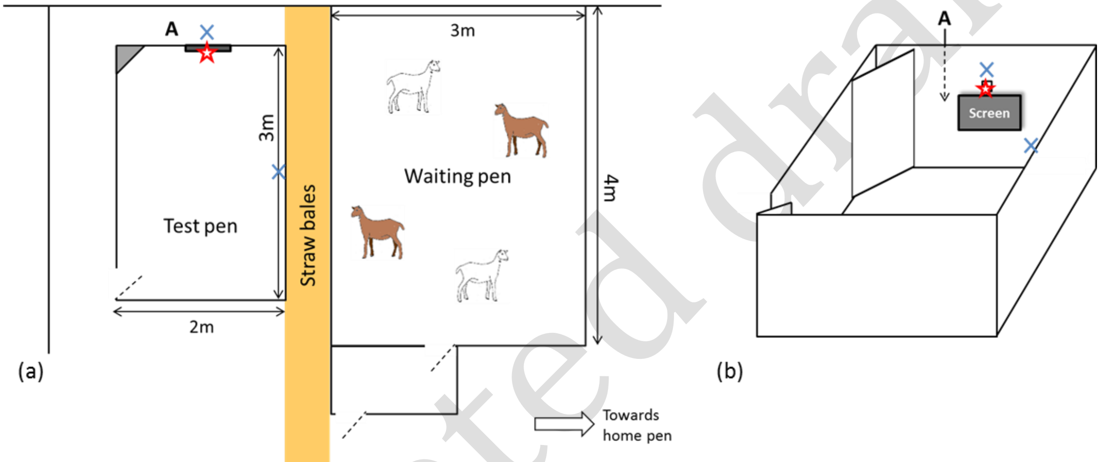

Figure 2. (a) Schematic representation of the test and waiting pens. (b) 3D view of the test pen. 'A' indicates the location of a hidden experimenter responsible for catching the goat's attention and the red star where the items were moved above the screen to catch the goat's attention. Blue crosses represent the two cameras.

2.4.2 Habituation

Goats were habituated to the experimental set-up prior to the beginning of the tests session. The habituation day was divided into three sessions. In the first session, the goats were brought into the test-pen in pairs once for 5 minutes. In the following two sessions, they were brought into the test-pen alone for 2 minutes.

2.4.3 Test sessions

The three days of testing followed the habituation. Order of testing was balanced as far as possible for breed and identity of the Photo Goat displayed as well as for the type of image. The order of testing of the goats was the same in all sessions. Each goat was shown the images of one Photo Goat of its own breed, resulting in the images of each Photo Goat being presented to a total of eight other goats.

Goats (including the Photo Goats) were exposed to one per test session, with two sessions per day and a total of six sessions across three days, with a different image shown in each session. For Sessions 1 and 2, the images shown were always the real positive and negative images, to obtain a baseline of the goats' reaction to images of real faces to allow comparison of the two emotions. In each group, for Session 1 half of the goats were exposed to the positive image and the other half to the negative. For Session 2, the goats were exposed to the second type of real image compared to Session 1. During Sessions 3, 4 and 5, the goats were exposed to the morphed images (I+, I50, I-). The order of testing of each morphed face was balanced so that in each session the same number of goats saw a given morphed face. Session 6 was a repetition of Session 1, and was used to test if the goats were still paying a similar level of attention to the image and if they were still reacting to the image.

One hour after morning milking, 16 goats were brought on a leash to the waiting pen. 15 minutes after the arrival of the last goat, the first goat was taken on a leash to the test pen. The test started when the door of the test pen was locked behind the goat. An image was displayed as soon as the goat paid attention to the (dark) screen. A goat fulfilled the 'paying attention' criterion when its head was oriented towards the screen for at least 1 second. To direct the attention of the goat towards the screen, an experimenter hidden behind the screen (position A) waved items through the opening made above the dark screen at the start of each session (Figure 2). The experimenter tried to catch the goat's attention until the goat fulfilled the criterion. There was no time limit, and it took 32 seconds on average, ranging from 0.2 to 216 seconds. Once this occurred, an image of a Photo Goat's face was displayed on the screen for 3 seconds after which the screen went dark again. We chose a presentation length of 3 seconds because we were only interested in the spontaneous reactions of the goats to the images. Limiting habituation to the presentation of images was also key due to the repeated exposures, and a very short exposure to the stimuli helped to preserve the goat's relative naivety towards images of faces.

The behaviour of the goat was video recorded from the start of the test session until the image disappeared. The animal was then returned to the waiting pen and the next goat brought for testing. Forty-five minutes after the last goat was tested in the first session of the day, the first was tested again to start the second session, resulting in an interval of approximately two hours between sessions for each goat.

2.5 Data collection and analysis

Behaviours described in Table 1 were scored from the video recording for each test session using The Observer 5.0 (Noldus Information Technology, Netherlands). Due to the very short duration of observations, video playback speed was slowed down by a factor of 10 for behavioural observations, and so every change in ear postures was recorded.

The outcome variables were percentages of time spent with the ears forward, backward, asymmetrical, and the percentage of time spent interacting with the screen (oriented towards and/or touching). Horizontal ear postures did not occur, so this behaviour was not included in the analyses. Time spent with the ears in forward,

Table 1. Recorded behaviours and transformation applied to outcome variables in goats when shown images of familiar conspecifics on a screen for 3 sec. Ears postures were adapted from Briefer et al, 2015.

Behaviour		Description	Unit	Transformation	Type
Beginning of the test		Once the door of the test pen was locked behind the goat	**---**		**---**
Latency to pay attention to the screen		Latency between the beginning of the test and the display of the image on the screen	sec (0.2 to 216 sec)	Ln	Predictor
Distance to the screen when image displayed		Estimated distance between the tip of the nose and the screen, when the image appears on the screen	6 categories from 50 – 300 cm	---	Predictor
Interacting with the screen		Time spent with the 2 eyes and the head in direction of the screen, regardless of the direction of the body (“looking”) or touching the screen (nose or lips touching the screen) while the image is displayed on the screen	sec (0.2 to 3 sec)	Logit	Outcome
Ear postures	**Ears forward**	Tip of both ears pointing towards the front of the goat	sec (0 to 3 sec)	Logit	Outcome
	Ears backward	Tip of both ears pointing towards the back of the goat	sec (0 to 3 sec)	Logit	Outcome
	Ears asymmetrical	Right and left ears in different position regarding a perpendicular to the head-rump axis	sec (0 to 3 sec)	Logit	Outcome
	Ears horizontal	Ears in a central posture, along a perpendicular to the head-rump axis	sec (0 to 3 sec)	Logit	Outcome

backward or asymmetrical postures summed to the total duration of the observation (sum-one constraint). Outcome variables were logit-transformed to conform to assumptions of the normality and homogeneity of the data. The predictor variable latency before the goat reached the 'attention OK' criterion (LatCrit) was ln-transformed for the same reasons. Time spent with the ears asymmetrical could not be transformed to conform with normality assumptions, and was thus transformed into a binomial variable (1 = asymmetrical ears occurred, 0 = asymmetrical ears did not occur).

All outcome variables were analysed for the 3 seconds interval when the image was displayed. Analyses were conducted in GenStat 16th edition (VSN International Ltd., United Kingdom). The significance level was set at P=0.05. All data in the text are presented as means ± 1 standard deviation, unless otherwise stated. We first compared spontaneous reactions to all five types of images, taking into account all six sessions. Continuous data were analysed by linear mixed models (REML) with repeated measurements. A power model for covariance was used to account for correlations within subjects across time. Power models allow unevenly spaced time points to be taken into account (e.g. that Sessions 1 and 2 were on the same day and thus closer in time than Sessions 2 and 3), since the correlations between measurements decrease as time between measurements increases. Heterogeneity of variance across test sessions was allowed when it led to a smaller deviance of the model (one-tailed test with a chi^2 distribution). The occurrence of the asymmetrical ear postures was analysed by general linear mixed model (GLMM) with a binomial distribution and logit link function. Breed, the type of image shown during the test session (TypeImage), Identity of the Photo Goat, the type of image shown during the previous session (PrevIm), DistScreen and LatCrit were considered as potential fixed effects. The interaction between TypeImage and Identity of the Photo Goat was also included in the list of potential fixed effects, since it was the most biologically relevant interaction in our design. Fixed effects were then fitted by stepwise backward selection for each outcome variable, and not all fixed effects listed above were included in the final model for each variable (see Table 2 for a detailed description of the fixed effects considered simultaneously in the final models). When a predictor was not included in the final fitted model, no statistical results are presented for that predictor. Session and Animal were included as random effects as Animal nested within Session. Post-hoc analyses were conducted using Fisher's Least Significant Difference tests. Data from Sessions 1 and 6 were also analysed separately following the same method, to compare the responses of the goats to the same stimuli presented twice.

Table 2. Final fixed effects fitted by stepwise backward selection for each outcome variables (forward, backward and asymmetrical ear postures and time spent interacting with the screen)

Variable	**Fitted fixed effects**
Forward	Breed + TypeIm[1] + iPG[2] + PrevIm[3] + DistIm[4] + DistScreen[5] + LatCrit[6]
Backward	Breed + TypeIm[1] + iPG[2] + PrevIm[3] + DistIm[4] + DistScreen[5]
Asymm.	Breed + TypeIm[1] + iPG[2] + TypeIm*iPG +DistScreen[5] + LatCrit[6]
Interacting	Breed + TypeIm[1] + iPG[2] + TypeIm.iPG + PrevIm[3] + DistScreen[5]

[1]TypeIm = Type of image displayed on the screen, could be positive, negative, I+ (75% positive), I50 (50% positive), and I- (25% positive)
[2] iPG = identity of the goat displayed on the screen (Photo Goat)
[3] PrevIm = type of previous image shown
[4] DistIm = relative distance to the previous image shown.
[5] DistScreen = Distance in cm between the head of the goat and the screen when the photo appeared
[6] LatCrit = latency before the goat reached the 'paying attention' criterion, i.e. stared at the screen for at least 1 sec

3 Results

3.1 Spontaneous reactions to different types of images

Goats reacted differently to different images of faces (Positive, I+, I50, I- and Negative). TypeImage had a significant effect on forward ear postures ($F_{4,121.3} = 2.51$, P = 0.045). Post-hoc comparisons showed that goats spent significantly more time with their ears forward when the negative image was shown compared to the positive image ($F_{4,121.3} = 2.51$, P = 0.018) (Figure 3a). There was no significant effect of TypeImage on time spent with the ears backward ($F_{4,139.4} = 1.73$, P = 0.147, Figure 3a) or on the occurrence of asymmetrical ears ($F_{4,53.9} = 0.34$, P = 0.850, Figure 3a).The interaction between TypeImage and the identity of the Photo Goat had an effect on time spent interacting with the screen ($F_{12,73} = 3.65$, P < 0.001, see Supplementary Information for more details).

Identity of the Photo Goat had an effect on the time spent with the ears forward ($F_{2,57.4} = 7.01$, P = 0.002) but not on the time spent with the ears backward ($F_{2,29.6} = 1.35$, P = 0.274, Figure 3b) or on the occurrence of asymmetrical ears ($F_{2,26.8} = 0.10$, P = 0.905, Figure 3b). Thus, goats exposed to images taken from Photo Goat 'Saanen 2' spent more time with their ears forward (Figure 3b) regardless of the type of image shown. Conversely, goats that looked at images taken from 'Alpine 2' spent less time with their ears forward (Figure 3b) compared to other Photo Goats except from 'Saanen 1', regardless of the type of image shown.

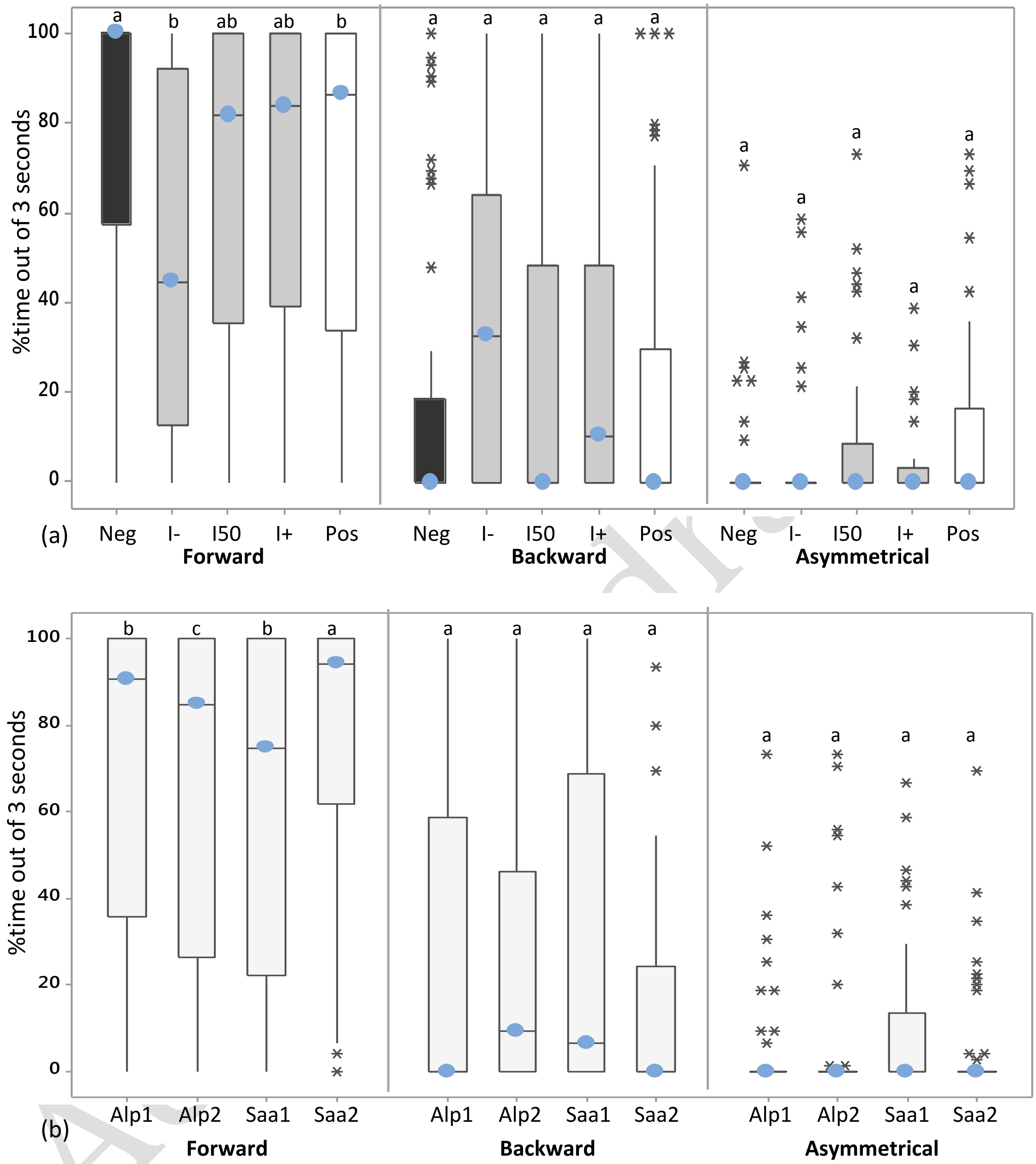

Figure 3. Effect of the type of images (a) and of the identity of the goat on the image (b) on the percentage of time spent in different ear postures in goats when shown different types of images of faces of familiar conspecifics on a screen for 3 sec.

Five images of the same goat of its own breed (Alpine (Alp) or Saanen (Saa)) were shown to any given goat. 'Negative': image taken while an icepack was applied to the goat udder. 'Positive': image taken while the goat was being groomed by a familiar experimenter. The three other types of images were of ambiguous facial expressions created using morphing software (25% positive (I-), 50% positive (I50), and 75% positive (I+)). P<0.05 when the bars share no common letters. Medians are indicated by a blue dot.

The distance between the goats and the screen when the image appeared (DistScreen) also affected the goats' ears postures in reaction to images (Figure 4). Regardless of the type of image shown, the further a goat was standing away from the screen when the image appeared, the more time the goat spent with its ears forward ($F_{5,145.4}$ = 10.22, P < 0.001). In contrast, the closer a goat stood from the screen, the more time it spent with the ears backward ($F_{5,165.3}$ = 7.89, P < 0.001, Figure 4) and the more asymmetrical ear postures it displayed ($F_{5,143.4}$ = 2.7, P = 0.019, Figure 4). DistScreen only tended to affect the total time spent interacting with the screen ($F_{5,91.9}$ = 2.08, P = 0.075).

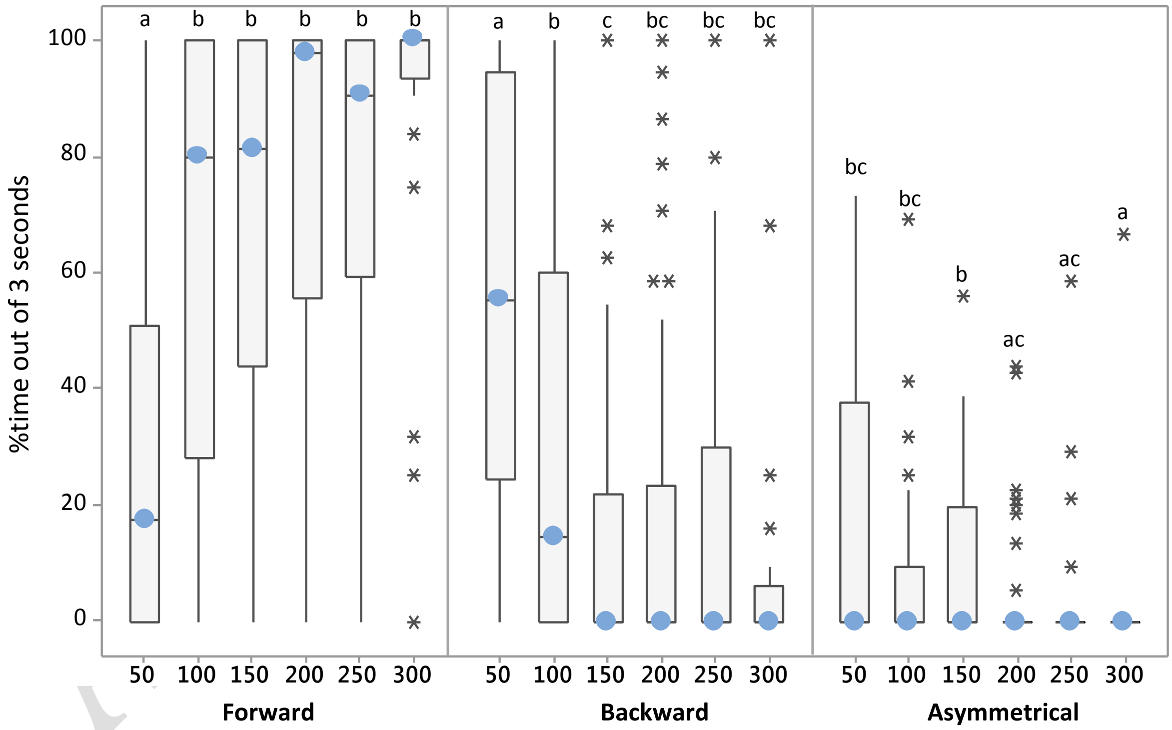

Figure 4. Effect of DistScreen, the estimated distance between the goat's head (tip of the nose) and the screen when the image appeared, on time spent with ears forward, backward and asymmetrical in 32 goats. DistScreen was divided into 6 categories, from 50 cm to 300 cm. P<0.05 when bars share no common letters. Medians are indicated by a blue dot.

Finally, Alpine goats spent longer interacting with the screen than Saanen goats ($F_{1,66.5}$ = 4.39, P = 0.040; Alpine: 75 ± 29%, Saanen: 68 ± 37%). There was no effect of breed on any of the other outcome variables (Ears Forward: $F_{1,54.8}$= 0.04, P = 0.836; Ears backward: $F_{1,28.4}$ = 0.14, P = 0.706; Ears asymmetrical: $F_{1,28.2}$ = 0.02, P = 0.793).

3.2 Repeated exposure to the stimuli

The type of previous image seen (PrevIm) had a significant effect on time spent interacting with the screen ($F_{5,86.8}$ = 11.54, P < 0.001) as well as on time spent in forward ($F_{5,111.6}$ = 2.96, P = 0.015) and backward ($F_{5,165.3}$ = 7.89, P < 0.001) ear postures (Figure 5). Post hoc analyses showed that this effect was due to the first session only, i.e. when there had been no previous image. Goats interacted with the screen for longer, spent more time with the ears forward and less time with the ears backward during the first Session than during any of the following sessions. PrevIm was not included in the final fitted model for the occurrence of asymmetrical ear postures.

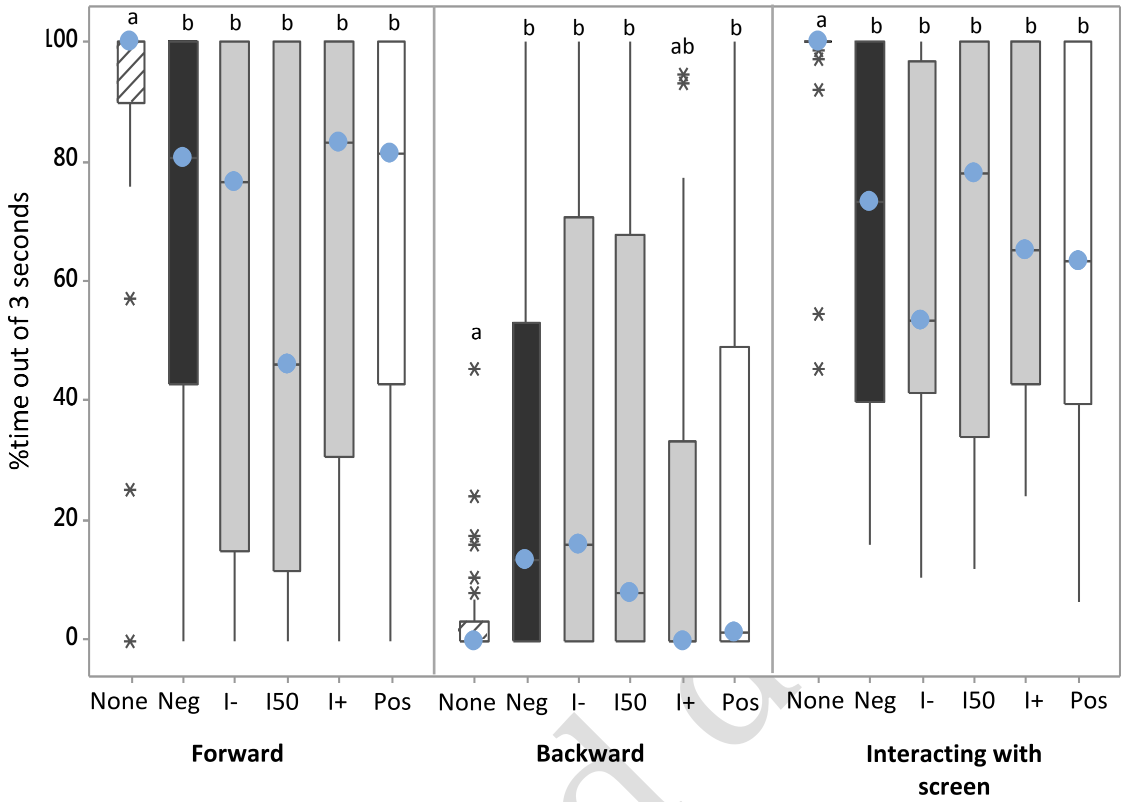

Figure 5. Effect of the type of previous image shown on the screen (PrevIm) on time spent with the ears forward and backward and on time spent interacting with the screen. None = no previous image (i.e the first test session), 'Neg': the previous image was the face of a goat taken while an icepack was applied to the udder. 'Pos': the previous image was the face of a goat taken while the goat was being groomed by a familiar experimenter. The three other types of previous images were of ambiguous facial expressions created using morphing software (25% positive (I-), 50% positive (I50), and 75% positive (I+)). P<0.05 when the bars share no common letters. Medians are indicated by a blue dot.

Session 6, as a repeat of the first session, allowed a check of the validity of the goats' response to images of faces after five repeated exposures. Goats spent more time with the ears in forward ear postures when negative images were shown in both sessions (Session 1: Positive = 82.2 ± 29.9%, Negative = 97.3 ± 6.9%, $F_{1,22}$ = 9.62, P=0.005 ; Session 6: Positive = 42.5 ± 42.1%, Negative = 61.7 ± 43.4%, $F_{1,19}$ = 7.60, P = 0.013). For the other ear postures, results in Session 6 were in the same direction as those in Session 1; however, those differences were not statistically significant. Time spent with the ears forward during Session 1 was correlated with time spent with the ears forward during Session 6 ($r_{p1\text{-}6}$ = 0.48, P<0.006). According to Martin and Bateson

(2007) this indicates a "substantial relationship" between Sessions 1 and 6 where the same image was shown (positive for some goats and negative for others). This relationship between Session 1 and 6 did not appear for other behavioural variables (ears backward: $r_{p1\text{-}6}$ = 0.22, P = 0.23; ears asymmetrical: $r_{p1\text{-}6}$ = 0.05, P = 0.79; time spent interacting with the screen: $r_{p1\text{-}6}$ = 0.18, P = 0.54).

4 Discussion

Differences in reactions to the different types of images

Our first hypothesis was that goats would show differences in their reactions to images of goats' faces taken in positive and negative situations, and that they would display behaviours indicating negative valence when looking at negative faces, and positive valence when looking at positive faces.

We found that goats displayed more ears forward when the image of a negative face was shown compared to a positive one. In sheep and goats, a higher percentage of time spent with the ears forward has been observed in situations with a negative valence situations, such as when the animal is being pricked by an experimental device (Vögeli et al., 2014) or when the animal is in socially isolated (goats: Briefer et al., 2015; sheep: Reefmann et al., 2009). However, a decrease in the percentage of time spent with the ears forward was observed after tail-docking and castration in lambs (Guesgen et al., 2016), suggesting that the association between ears forward cannot be generalised to all negatively valenced situations. In fact, forward ear postures have also been observed in situations where a high level of attention is required, i.e. eliciting high arousal (exposure to an unfamiliar test situation involving mild pain in sheep (Stubsjøen et al., 2009), or novel odour test in wild mice (Lecorps and Féron, 2015)).

Situations eliciting high arousal often coincide with a negative valence, but empirical observations have also identified forward ear postures in what could be considered positive situations, for instance, while the animals approached rapidly a bucket containing food pellets or when a familiar human entered the barn (personal observations). A higher percentage of time spent with the ears forward could then be associated with situations that lead to high arousal and/or increased attention, rather than to negative situations *per se*. Since most negative situations lead to an increase in attention to the environment (Carretié et al., 2001), this would explain the repeated occurrence of higher proportions of forward ear postures in negative situations.

Different situations can induce similar emotional states and facial expressions (including ear postures). For instance, social isolation (Briefer et al., 2015) and pain caused by castration and tail docking (Guesgen et al., 2016), both negative situations, have been associated with backward ear postures in small ruminants. It can thus be consider that here, goats perceived the valence of the situations as being positive or negative, rather than specificities of the situation, e.g. pleasurable handling or discomfort to the udder. To rule out alternative explanations would require repeating these tests with images taken in two different positive and negative situations. Based on our results, images of faces taken during a negative situation seem to have elicited higher attention and arousal amongst the tested goats. This might indicate that images of faces taken during a negative situation were perceived as more negative stimuli by the goats, or at least elicited more attention than images of faces taken during a positive situation. From a behavioural ecology point of view, it is appropriate for prey animals such as goats to pay more attention to faces displaying negative emotions as they could signal the presence of potential threats. The association of forward ear postures and increased attention in goats is further supported by the fact that the further a goat stood from the screen, the more time it spent with its ears forward. This could indicate that the animal was directing its attention towards the screen while keeping a safe distance, thus displaying higher alertness. The lower proportion of ears forward observed when a positive image was shown would then indicate that goats were less attentive, and that the goats could have perceived images of faces taken during a positive situation as more positive or as less interesting.

In our study the percentage of time spent in backward ear postures was fairly low (20% on average), which represents an actual duration of less than 1 second. As such our results need to be treated with caution. The percentages of time spent with asymmetrical postures were even lower (≤10% on average), which is in agreement with observations made by Briefer et al. (2015). As pointed out by Guesgen et al (2016), although discrete ear postures were analysed, those postures were mutually exclusive. In other words, if the proportion of time spent with the ears forward decreased, the proportion of time spent in other ear postures increased. This could be another explanation for the higher percentage of time spent in asymmetrical ear postures that we observed when a positive image was shown. The three types of ear postures we recorded were indeed not independent and thus should be interpreted simultaneously. However, to identify how a situation was perceived, it is not the changes in ear postures, but rather the direction of the change (higher proportion of ears forward for instance) that is of interest.

Overall, these differences in ear postures indicated that goats paid more attention to images of conspecifics in a negative situation than to images of conspecifics in a positive situation. The fact that goats are able to identify faces of conspecifics in a negative situation, and so potentially a negative emotional state, could have welfare implications. From that perspective, it would be interesting to assess the impact of such images on the emotional state of the goat that is observing them. This, in the long term it might lead to a better understanding of the impact of seeing conspecifics in a negative emotional state and its implication from a welfare point of view.

Potential use of images of faces in cognitive bias studies

If our results indicate that goats can discriminate between images of faces displaying different facial expressions, it is still unclear which facial features were indicative of the valence of the situation. Based on previous studies in sheep (Peirce et al., 2000; Tate et al., 2006), it is reasonable to assume that ear postures were important cues that the goats used to discriminate between images of faces. Variations in ear postures were also the most visible difference between the different types of images. Additional studies using modified images hiding specific facial features (eyes, ears, mouth) would allow us to test this hypothesis (Wathan and McComb, 2014).

Our second hypothesis was that goats would show reactions to the morphed faces that were intermediate to the negative and positive images, and would reflect a gradual change in their response to the images, from the more negative to more positive image. However, the responses to morphed faces we observed were not necessarily intermediate for all behaviours (ears backwards and ears asymmetrical especially) and the variation in responses to morphed images was not gradual. While this result is not encouraging regarding the use of images of faces for cognitive bias studies, it is worth noting that responses to the two extreme cues agreed with our hypotheses, and that difficulties arose with the morphed images. Morphed images have been used successfully as ambiguous stimuli in previous judgment bias studies in chickens (Salmeto et al., 2011), but they consisted of silhouettes of birds and not complex stimuli such as faces. Further work is thus needed to better understand how morphed images of faces are perceived by goats, and which facial features matter most for face-based perception of emotions.

Methodological limitations

The identity of the Photo Goat affected the spontaneous reaction of goats to the images. This did not affect the general direction of the results, but it did affect more the magnitude of the responses. For instance goats tested with images of Photo Goat 'Saanen 2' displayed more forward ear postures overall, while still following the general response of a higher percentage of time spent with the ears forward when a negative face was shown. Dominance relationships, but also affinity between the tested and photographed individual, could have affected the goats' responses.

In this study, we presented the same image of a given Photo Goat in given situation to test goats. As a future refinement to this methodology, it would be important to understand whether a series of separate images of a given Photo Goat in a particular emotional state are perceived similarly within and between test animals. However, this study was a first step in investigating face-based emotion-recognition in goats, and allowed us to assess the effect of the type of image presented, with a satisfactory degree a generalisation (four different images were presented for each type of image). More studies would be needed, with experimental designs involving more images to determine how general these responses are.

We saw a strong effect of session on behaviour. Specifically, there was a difference between the first session and the other sessions in interest and attention. The higher interest for the image shown during the first session could be due to a novelty effect that quickly faded (Désiré et al., 2004). However, even though the percentage of time spent interacting with the screen dropped after the first session it stayed above 60% until the last session. In fact, even after five exposures to the stimuli, the goats still paid attention to the image.

Finally, the group of animals included in this study was as homogeneous as possible, especially in terms of age and previous experience. These factors may affect goats' responses. Further research would be thus be needed to clarify this point, since there is a possibility that this group differed from the general population for various reasons, including for example their past experience, the location of their home pen, the influence of one specific group member on the other animals, and difference in their relationships with humans. Choosing to include goats housed in separate pens, of varying ages and experiences could have lead to more generalizable results. However to avoid confounds between individual characteristics and, for instance, the type of image shown, we chose to study a homogeneous group.

5 Conclusion

Goats showed different reactions to images of faces photographed in different situations, indicating that they perceived the images as different. Goats also appeared more attentive towards negative images than towards positive or morphed images, which could indicate that negative images were, in fact, perceived to be more. Responses to morphed images were not necessarily intermediate to responses to negative and positive images and not gradual either, suggesting that using images of faces in cognitive bias tests may be inappropriate. Further study of the perception of morphed faces is needed. In addition, future research should take into account the fact that the goats appeared to be sensitive to the novelty of the stimulus and the identity of the individual in the photo.

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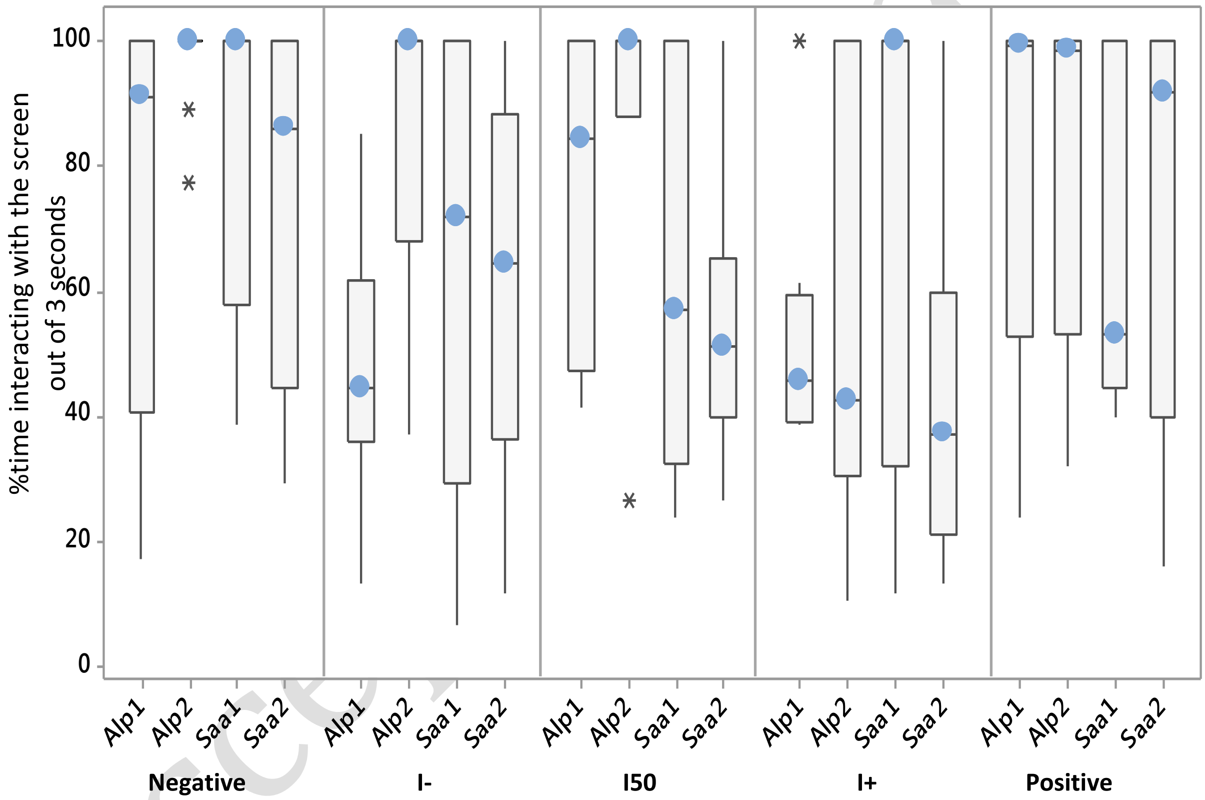
7 Supplementary information

Results from post hoc analyses (Fishers' least significant difference tests) showing the effect of the interaction between the identity of the Photo Goat and the type of image shown on the percentage of time spent interacting with the screen. The overall effect of the interaction was $F_{12,84.7} = 3.02$, P = 0.001.



Supplementary Figure 1. Effect of the interaction between the identity of the Photo Goat and the type of image shown. Medians are indicated by a blue dot. 'Negative': image taken while an icepack was applied to the goat udder. 'Positive': image taken while the goat was being groomed by a familiar experimenter. The three other types of images were of ambiguous facial expressions created using morphing software (25% positive (I-), 50% positive (I50), and 75% positive (I+))

Supplementary Table 1. P-values for the effect of the interaction between the identity of the Photo Goat and the type of image shown on the percentage of time spent interacting with the screen. For readability reasons, the second half of the table has not been filled symmetrically. Significant differences ($P < 0.05$) are indicated in bold text in Table (b).

		Neg.				I-				I50				I+				Pos.			
		Saa1	Saa2	Alp1	Alp2	Saa1	Saa2	Alp1	Alp2	Saa1	Saa2	Alp1	Alp2	Saa1	Saa2	Alp1	Alp2	Saa1	Saa2	Alp1	Alp2
Neg.	Saa1																				
	Saa2	0.55																			
	Alp1	0.17	0.09																		
	Alp2	0.56	0.37	0.13																	
I-	Saa1	0.82	0.84	0.17	0.51																
	Saa2	0.10	0.04	0.96	0.53	0.09															
	Alp1	**0.02**	**0.01**	0.08	**0.01**	**0.02**	0.25														
	Alp2	0.99	0.80	0.10	0.47	0.90	0.30	**0.003**													
I50	Saa1	0.87	0.56	0.11	0.57	0.74	0.21	**0.01**	0.90												
	Saa2	0.40	0.21	0.52	0.96	0.39	0.47	0.11	0.63	0.54											
	Alp1	0.87	0.56	0.11	0.57	0.74	0.21	**0.01**	0.90	0.47	0.54										
	Alp2	0.61	0.81	**0.02**	0.16	0.73	0.15	**0.001**	0.57	0.44	0.32	0.44									
I+	Saa1	0.06	0.16	**0.02**	0.09	0.18	**0.005**	**0.002**	0.32	0.09	**0.03**	0.09	0.57								
	Saa2	0.13	0.06	0.90	0.59	0.15	0.32	0.25	0.93	0.25	0.54	0.25	0.18	**0.004**							
	Alp1	**0.02**	**0.01**	0.10	**0.01**	**0.03**	0.31	0.95	**0.01**	**0.01**	0.12	**0.01**	**0.002**	**0.002**	0.26						
	Alp2	**0.02**	**0.01**	0.10	**0.01**	**0.03**	0.29	0.97	**0.009**	**0.01**	0.12	**0.01**	**0.002**	**0.001**	0.24	0.97					
Pos.	Saa1	**0.01**	**0.002**	0.91	0.50	**0.05**	0.94	0.20	0.29	0.12	0.41	0.12	0.12	**<0.001**	0.97	0.22	0.21				
	Saa2	0.39	0.16	0.35	0.99	0.42	0.27	**0.05**	0.71	0.68	0.80	0.68	0.39	**0.01**	0.31	0.06	**0.05**	0.09			
	Alp1	0.17	0.09	0.98	0.12	0.17	0.95	0.12	0.10	0.11	0.52	0.11	**0.02**	**0.02**	0.90	0.12	0.10	0.90	0.34		
	Alp2	0.15	0.09	0.87	0.12	0.16	0.90	0.08	0.09	0.11	0.48	0.11	**0.02**	**0.02**	0.84	0.14	0.13	0.84	0.32	0.89	

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